# ANOPHELES (CELLIA) AINSHAMSI, N. SP. (DIPTERA: CULICIDAE), A SALTWATER SPECIES FROM THE RED SEA COAST OF EGYPT 

Adel M. Gad, Ralph E. Harbach, and Bruce A. Harrison

(AMG) Department of Entomology, Faculty of Science, Ain Shams University, Abbassia, Cairo, Egypt (e-mail: adelgad2002@yahoo.com); (REH) Department of Entomology, The Natural History Museum, Cromwell Road, London SW7 5BD, U.K. (e-mail: r.harbach@nhm.ac.uk); (BAH) North Carolina Department of Environment and Natural Resources, Public Health Pest Management, 585 Waughtown Street, Winston-Salem, NC 27107, U.S.A. (e-mail: bruce.harrison@ncmail.net)

Abstract.-The Anopheles species that breeds in saltwater pools on the Red Sea coast of Egypt, originally thought to be An. stephensi (Liston), is formally described and named Anopheles (Cellia) ainshamsi, n. sp. The adults, pupa, and fourth-instar larva are characterized, and the male genitalia and the two immature stages are illustrated. The species is compared and distinguished from similar members of the Neocellia Series that occur in southwestern Asia and Africa. Attempts to obtain DNA sequence data from type specimens and other specimens collected more than 20 years ago were unsuccessful.

Key Words: Anopheles ainshamsi, dancalicus, hervyi, salbaii, stephensi, mosquitoes, new species, Neocellia Series

Gad (1967) found Anopheles larvae breeding in saltwater pools near the Red Sea coast of Egypt that he identified as An. stephensi (Liston). Gad realized that An. stephensi was not known to occur in Africa or western areas of the Arabian Peninsula, and sent specimens to the British Museum (Natural History) (now known as The Natural History Museum) to confirm his identification. The late Peter Mattingly examined the specimens and noted: "The wing is rather paler than usual. The leaflets on the phallosome are rather smaller than figured by Christophers for stephensi . . . . The most striking difference in the larvae seems to be quite heavy spiculation of the inner, and sometimes outer clypeal hairs [setae 2C and 3-C, respectively], and the branching of the post clypeal [seta 4-C]. .." (Gad and Kamel 1967). Despite these differences, it
apparently never occurred to either Gad or Mattingly that the species might not be An. stephensi.

In 1981, one of us (AMG, and colleagues) established a colony of this mosquito to study its biology. Unlike typical An. stephensi, females of the Egyptian mosquito were autogenous, i.e., they laid eggs without taking a blood meal (documented by Ribeiro et al. 1985). This prompted AMG to compare specimens from the Red Sea coast with unequivocal specimens of An. stephensi (Rangoon strain) from a previous study (Gad et al. 1979a, b). Specimens from Egypt were found to differ significantly from typical An. stephensi, and to share certain characteristics with two other African anophelines, An. dancalicus (Corradetti) from the Danakil Depression of Ethiopia and An. salbaii (Maffi and Coluz-
zi) from the Ogaden Desert of Somalia. It also bears similarities with An. hervyi Brunhes, le Goff, and Geoffroy, which was recently described from adult females found in the Sahelian area of southern Niger (Brunhes et al. 1999). Accordingly, the Egyptian species, known informally as An. ainshamsi for nearly 25 years, is formally described and named An. ainshamsi, n. sp. in this report.

## Materials and Methods

This study is based on specimens collected near Râs Shukeir on the Red Sea coast (Gulf of Suez) of Egypt in 1983. Wild-caught larvae were reared individually in water from their natural breeding sites to obtain adults with associated larval and pupal exuviae. Some fourth-instar larvae were also preserved for study. Comparisons were made with specimens of similar species deposited in The Natural History Museum (NHM), London, including type specimens of An. dancalicus, An. hervyi, and An. salbaii. Observations of the adults were made under simulated natural light. Larval and pupal chaetotaxy were studied using differential interference contrast microscopy. Measurements and counts, except for structures of the male genitalia, were taken from $10-15$ specimens. Numbers in parentheses represent modes of the reported ranges. Except for wing spot nomenclature (Wilkerson and Peyton 1990), the morphological terminology used in the species description follows Harbach and Knight (1980, 1982). Recognition of the new species is based on diagnostic and differential characters that distinguish it from its closest allies. The symbols $\mathrm{A}, \stackrel{\circ}{ }$, ${ }^{\circ}, \mathrm{Le}, \mathrm{Pe}$, and L used in the literature summary and material examined sections represent adult, female, male, larval exuviae, pupal exuviae, and fourth-instar larva, respectively. An asterisk ${ }^{(*)}$ after one of these symbols in the literature summary section indicates at least part of the life stage was illustrated in the publication cited. Type specimens are deposited in the National Museum of Natural

History (NMNH), Washington, DC, U.S.A., Ain Shams University (AU), Cairo, Egypt, and the NHM, London, U.K.

## Taxonomic Treatment

## Anopheles (Cellia) ainshamsi Gad, Harbach, and Harrison, n. sp.

 (Figs. 1, 2)Anopheles ainshamsi (nomen nudum) of Gad et al. 1987: 211, 214, 217 (L bionomics); Ward 1992: 210 (catalog).
Anopheles sp. nr. salbaii of Ribeiro et al. 1985: 689-692 (A biology).
Anopheles stephensi of Gad 1967: 172-174 (L*, L bionomics); Gad and Kamel 1967: 249-252 (L bionomics, A, L morphology); Gad and Salit 1972: 581 (A biology); El-Said and Kenawy 1983: 69, 73 (collection record); Kenawy 1988: 74 (bionomics, distribution); Kenawy 1990: 270, 271 (collection record).
Anopheles (Cellia) n. sp. of Glick 1992: 129, 130, 140, 150 (distribution, A key).

Diagnosis.-Anopheles ainshamsi is a member of the Neocellia Series based on the absence of upper proepisternal setae in adults and the presence of one branched long propleural, one branched long mesopleural, and two branched long metapleural setae (9-P, 9-M, and 9,10-T, respectively) in larvae. Adults of An. ainshamsi are distinguished from Oriental members of the series, except An. stephensi, in having both spotted legs and hindtarsomere 5 darkscaled, and from Afrotropical members of the series, except An. dancalicus, An. herv$y i$, and An. salbaii, by the combination of spotted legs, dark hindtarsomere 5, and scaling on abdominal segments II-VIII. The absence of scales on the scutal fossa distinguishes An. ainshamsi from An. stephensi and the three similar Afrotropical species. Anopheles ainshamsi resembles An. dancalicus and differs from the other three species in lacking a pale fringe spot at the apex of the anal vein of the wings. Larvae are distinguished from other mem-

ber of the series by one or more of the following characters: seta 2-C not brush-like; 2,3-C aciculate or frayed; 1,2-P well separated, inserted on small tubercles; 2-P with fewer than 15 branches; 11-P without short barblike branches; 1-III-VII lanceolate branches or blades with weakly developed shoulders and a short filament. Pupae of species belonging to the series are not well known and are difficult to distinguish.

Female.-Overtly brown and pale yellow. Head: Truncate erect scales of vertex pale (white) anteriorly and becoming progressively darker (yellowish to brown) posteriorly and posterolaterally; eyes widely spaced, erect scales grade into elongate semierect fusiform scales on interocular space, these scales interspersed with long golden setae, lateral margins of interocular space lined with white decumbent scales that become longer and give rise to long sinuous setae above antennal pedicels. Clypeus bare. Antenna length $0.9-1.2 \mathrm{~mm}$ (mean 1.0 mm ); pedicel with yellowish to brown integument and usually few inconspicuous pale scales on dorsal surface; flagellomeres 1-3 with elongate pale scales, particularly dense on mesal surfaces. Proboscis $1.4-1.7 \mathrm{~mm}$ (mean 1.6 mm ), slightly longer than forefemur (about $1.1 \times$ ); prementum entirely dark-scaled, scales appressed throughout except for few slightly erect scales at base; labella slightly paler than prementum. Maxillary palpus length $1.4-1.6 \mathrm{~mm}$ (mean 1.5 mm ), usually with 3 pale (white) bands-apical (apex of palpomere 4 and all of 5), preapical (apex of 3 and base of 4) and proximal (apex of 2)apical pale band about length of preapical dark band (middle of palpomere 4), palpus with 4-banded appearance when middle of palpomere 5 occasionally dark-scaled; palpomere 2 with semi-erect scales giving a slightly bushy appearance to proximal portion of palpus; ventral surface of palpus without scales. Thorax: Integument dark brown; scutum with broad median pale pruinose area confluent with scutellum of similar appearance; anterior promontory and
antedorsocentral areas with white semierect scales that grade into yellowish to golden decumbent fusiform scales on acrostichal and dorsocentral areas, scales on these areas converge at middle of scutum and extend posteriorly between posterior dorsocentral and lateral prescutellar setae to scutellum, with small posterior medial area of prescutellar area void of scales, scales at lateral margins of this bare area become white before margin of scutellum; golden to goldenbrown setae on acrostichal, dorsocentral, fossal, antealar, supraalar and prescutal areas; narrow line of decumbent pale scales on mesal side of supraalar setae extends to near parascutellar seta, this line of scales separated from scales on posterior dorsocentral and prescutellar areas by rather broad pale pruinose area. Scutellum with row of white to golden fusiform scales adjacent to posterior row of long goldenbrown setae. Mesopostnotum and postpronotum bare. Antepronotum without scales, with long golden-brown setae. Pleura with golden-brown setae: $0-4(1)$ prespiracular area, 1-3(2) prealar, 2,3(2) upper and 13(2) lower mesokatepisternal and 3-6(4) upper mesepimeral; upper proepisternal setae absent. Wing (see Fig. 62 in Glick 1992): Length 2.7-3.3 mm (mean 2.9 mm ); dark scaling brown, stark on costa, subcosta and $\mathrm{R}-\mathrm{R}_{1}$, subdued on posterior veins, pale scaling pale yellow, not white; costa with humeral dark spot, dark basal to humeral crossvein; costa, subcosta and $R$ with presector and sector dark spots, presector equally long on 3 veins, sector about half as long on R , sometimes with pale interruption; costa and $R_{1}$ with equally long preapical and apical dark spots; remigium and base of R pale to presector dark spot; dark spots on other veins often faint (some sometimes absent, fully present as follows: Rs dark except at base, $R_{2}$ with dark spot opposite apical dark spot on $\mathrm{R}_{2}$, spur of $\mathrm{R}_{4+5}$ dark, postbasal and preapical dark spots on $\mathbf{R}_{4+5}, \mathbf{M}_{1}, \mathbf{M}_{2}, \mathbf{M}_{3+4}$ and CuP, distal areas of M and mou dark-scaled, 1 A with 2 dark spots in distal half (preapical and

Table 1. Range (mode) of numbers of branches for pupal setae of Anopheles (Cellia) ainshamsi.

| $\begin{aligned} & \text { Seta } \\ & \text { No. } \end{aligned}$ | Cephalothorax CT | Abdominal Segments |  |  |  |  |  |  |  |  | $\begin{gathered} \text { Paddle } \\ \mathrm{Pa} \end{gathered}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | 1 | II | III | IV | v | Vi | VII | VIII | IX |  |
| 0 | - | - | 1 | 1 | 1 | 1 | 1 | 1 | 1 | - | - |
| 1 | 2-4 (3) | $\approx 80$ | 5-10 (8) | 4-8 (6) | 3-10 (4) | 1-4 (2) | 1,2 (1) | 1,2 (1) | - | 2, 3 (3) | 1 |
| 2 | 2-4 (2) | 2-8 (5) | 4-9 (5) | 3-7 (4) | 3-5 (3) | 3-5 (3) | 3-5 (3) | 2-5 (3) | - | - | 2-5 (4) |
| 3 | 3-5 (3) | 1,2 (1) | 1-4 (2) | 1-4 (3) | 2-7 (5) | 1-4 (1) | 1,2 (1) | 1-4 (2) | - | - | - |
| 4 | 2-5 (3) | 2-6 (4) | 1-7 (3) | 2-7 (5) | 3-6 (4) | 1-5 (4) | 1-3 (2) | 1,2 (1) | 1-3 (2) | - | - |
| 5 | +9(7) | 1-6 (5) | 2-6 (3) | 5-8 (6) | 2-8 (4) | 3-7 (3) | 2-5 (4) | 2-6 (4) | - | - | - |
| 6 | 2-5 (3) | 1-4 (3) | 3-5 (3) | 3-8 (5) | 3-7 (4) | 2-5 (3) | 2-4 (3) | $1-3(2)$ | - | - | - |
| 7 | 2, 3 (2) | 3-6 (4) | 2-6 (3) | 3-6 (4) | 3-6 (5) | 2-4 (3) | 1,2 (1) | $1-3$ (1) | - | - | - |
| 8 | 1,2 (2) | - | a | 2-5 (4) | 1-4 (3) | 1-4 (2) | 1-4 (3) | 1-3 (3) | - | - | - |
| 9 | 1-3 (2) | 1,2 (1) | 1 | 1 | 1 | 1 | 1 | 1-3 (1) | 7-11 (8) | - | - |
| 10 | 1-4 (2) | - | - | 2-5 (3) | 1-3 (1) | 1,2 (1) | 1-3 (2) | $1-3$ (2) | - | - | - |
| 11 | 1-5 (4) | - | - | 1,2 (1) | 1,2 (1) | 1,2 (1) | 1 | 1-4 (2) | - | - | - |
| 12 | 1-4 (2) | - | - | - | - | - | - | - | - | - | - |
| 14 | - | - | - | 1 | 1 | 1 | 1 | 1 | 1 | - |  |

$a=$ alveolus only.
just beyond midlength), apex of vein without scales; faint pale fringe spots at apices of $\mathbf{M}_{2}, \mathrm{M}_{3}, \mathrm{M}_{3+4}$ and CuP (total of 4 pale fringe spots). Halter: Pedicel and scabellum pale, capitellum dark-scaled. Legs: Forecoxa with few inconspicuous dark scales among setae anteriorly at base, midcoxa usually with few inconspicuous pale scales among setae laterally at base, hindcoxa without scales; femora, tibiae and first tarsomeres with speckles and blotches of pale yellow scaling; foretarsus with pale bands across joints, mid- and hindtarsi with narrow pale bands or dorsal pale spots at apices of tarsomeres $1-4$, sometimes faint or absent on midtarsomeres $2-4$, hindtarsomere 5 usually narrowly pale at base. $A b$ domen: Integument dark, with golden setae; tergum I and sterna I-VI without scales, terga II-VIII largely covered (except laterally) with pale yellow to golden fusiform and narrow spatulate (primarily) scales, cerci with similar scales, sternum VIII, and sometimes posterior area of sternum VII, with scattered pale scales (see Glick 1992: Fig. 64).

Male.-Similar to female except for obvious sexual differences; other differences include the following. Head: Proboscis
slightly longer, $1.6-1.8 \mathrm{~mm}$ (mean 1.7 mm ), about 1.6 length of forefemur. Maxillary palpus largely pale-scaled, dark scaling on palpomere 1 , proximal 0.5 or less of palpomere 2 and narrowly across joints between palpomeres $2-3,3-4$ and $4-5$. Wing: Length $2.5-3.0 \mathrm{~mm}$ (mean 2.7 mm ); generally paler and scaling reduced, dark spots of posterior veins very faint or absent, fringe spots unapparent. Genitalia (Fig. 1C): Gonocoxite with pale yellow scales on lateral surface; with 5 parabasal setae, most sternocaudal seta long and slender, similar to unspecialized setae of gonocoxite; gonostylus strongly and evenly curved in distal half, with row of minute setae along sternomesal margin and 1 or 2 longer setae on tergal side near apex; claspette with long apical seta about 1.5 length of club and 3 or 4 shorter subapical setae, club formed of 4 fused setae; aedeagus with 2 or 3 pairs of smooth, slender, attenuated leaflets; proctiger membranous, with lightly sclerotized long narrow lateral paraprocts.

Pupa (Fig. 1A, B).-Character and positions of setae as figured; numbers of branches in Table 1. Cephalothorax: Lightly tanned, legs darker, scutum and metanotum with darker blotches. Seta 7-CT
about 1.8 length of 6-CT, usually double, sometimes triple; 8 -CT normally single, rarely double; $10-\mathrm{CT}$ usual double or triple ( $1-4$ branches) with branches arising near base; 11-CT usually split distally into $2-5$ short branches, sometimes single. Trumpet: Angusticorn, moderately tanned, borne on tubercle, tracheoid absent, pinna without slit; length $0.37-0.46 \mathrm{~mm}$ (mean 0.42 mm ); meatus fairly long, $0.15-0.22 \mathrm{~mm}$ (mean 0.18 mm ); pinna slightly longer, $0.19-0.25$ mm (mean 0.23 mm ). Abdomen: Length 2.22-3.04 mm (mean 2.54 mm ); lightly tanned, anterior margins of sterna darker, progressively lighter after sternum IV. Seta 0-II-VII minute, simple, inserted anterior and usually slightly mesad of seta 2 ; seta $1-$ II-IV with multiple thin flexible branches, 1-V usually double ( $1-4$ branches) and longer than following tergum, 1-VI,VII usually single (infrequently double) and longer than following tergum; seta 6-II generally triple (3-5 branches) and nearly twice length of seta 7, 6-III-VII multiple branched, number of branches generally progressively decrease from 6-III to 6-VII; seta $7-\mathrm{III}$,IV often inserted within striations of fold line, $7-$ V-VII always inserted on fold line, 7-VII inserted at posterior margin of segment, seta 7-III-V short, branched, 7-VI,VII usually single, long, about length of following sternum: setae $8,10,11$-II absent, alveolus of 8 -II usually present; seta $9-\mathrm{I}$ relatively short, about 0.35 length of 6-I, usually single, occasionally double; 9-II-IV small, peg-like; 9-V-VII long, curved, simple and sharply pointed, length not substantially increasing from segment V to segment VII; 9-VIII plumose with 7-11(8) branches arising from a normally thickened non-flattened central stem. Genital lobe: Length about 0.25 mm in female; about 0.45 mm in male, with nipple at apex. Paddle: Lightly pigmented (hyaline), buttress and midrib slightly darker, midrib distinct to near seta $2-\mathrm{Pa}$; length $0.64-0.77 \mathrm{~mm}$ (mean 0.71 mm ), width $0.43-0.56 \mathrm{~mm}$ (mean 0.50 mm ), index 1.35-1.48 (mean 1.42); outer part with spicules ending before seta $1-\mathrm{Pa}$,
marginal serrations (refractile border) begin $0.26-0.40$ from base and end $0.48-0.66$ from base where they grade into short hyaline filaments; refractile index 0.25-0.43 (mean 0.35). Seta 1-Pa long, sinuous, with hooked tip, about one-third length of paddle; 2-Pa well developed, relatively long, with $2-5(4)$ branches.

Larva, fourth-instar (Fig. 2).-Character and positions of setae as figured; numbers of branches in Table 2. Head: Length $0.60-$ 0.72 mm (mean 0.67 mm ), width $0.64-0.77$ mm (mean 0.71 mm ); moderately tanned, darker patches behind setae 5-7-C and posterior to eyes, collar and dorsomentum darkly tanned. Seta 2-C single, aciculate or frayed in distal half; 3-C generally single but often with aciculae or dendritic processes; 4-C single, rarely double, relatively long, extending well beyond base of $2-\mathrm{C}$; $6,7-\mathrm{C}$ relatively short, about half length of 5-C, 11-C with comparatively few branches (8-18, commonly 15). Antenna: Moderately tanned; mesal and ventral surfaces with relatively sparse needle-like spicules; length $0.20-0.28 \mathrm{~mm}$ (mean 0.25 mm ). Seta $1-\mathrm{A}$ small, about as long as diameter of antenna, single, inserted about one-third distance between base and apex of antenna. Thorax: Integument hyaline, smooth. Seta 1-P on small setal support plate, with 5-8(7) rather widely spaced branches; 2-P on margin of plate bearing seta $3-\mathrm{C}$, single; 11-P significantly larger than $11-\mathrm{M}, \mathrm{T}$, with $2-4(3)$ branches; support plate of pleural setal group 9-12-P with small lateral spine, 9$\mathrm{P}, \mathrm{M}, \mathrm{T}$ and $10-\mathrm{T}$ always branched, $10-\mathrm{M}, \mathrm{T}$ and $12-\mathrm{P}, \mathrm{M}, \mathrm{T}$ often single but sometimes with 2 or 3 branches, 12-T more often triple; 14-P with relatively few branches (25 , usually 3 ); $4-\mathrm{M}$ usually with 2 or 3 branches arising from short stem, occasionally with 4 branches, rarely single or with 5 branches; 6-M rather long, usually with 3 or 4 branches arising from short stem, range 3-8 branches; 7-M farther ventrad of $6-\mathrm{M}$ than usual (not evident in Fig. 2), with 36(4) branches arising from short stem; 3-T very often single, sometimes with 2 or 3
PROCEEDINGS OF THE ENTOMOLOGICAL SOCIETY OF WASHINGTON
－рәииол юии $=$ จи



| － | － | －－ | － | － | － | － | － | － | － | － | － | ou | SI |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| － | I | I | 1 | I | 1 | I | － | － | － | （9） $8^{-\square}$ | （ $\downarrow$ ）$\varsigma-\varepsilon$ | ขu | ¢ |
| － | － | （ع）$L-\mathcal{E}$ | （9） $8-\varepsilon$ | （ع）$\varsigma-\varepsilon$ | （b）$\varsigma-\varepsilon$ | （¢）$\varsigma-\varepsilon$ | （c）9－ヤ | （t） $9-\varepsilon$ | （ع）$\nabla^{\prime} \varepsilon$ | （9） $6-S$ | （ع）$\varsigma-Z$ | （ع）$s-\tau$ | $\varepsilon I$ |
| （z）$\varepsilon-1$ | ＇S－6 | （乙）$\varepsilon$＇乙 | （乙）$\varepsilon-1$ | （z）$s-z$ | （z）$\varepsilon-1$ | （乙）$\varepsilon-1$ | （Z） $\mathcal{E}-1$ | （ع）$\varsigma-\tau$ | （E） $\mathcal{E}-1$ | （I） $\mathcal{E}-\mathrm{I}$ | （I）$\varepsilon-I$ | （b）$s-\tau$ | て1 |
| （t）$s-\tau$ | ＇S－8 | （て）$\downarrow-1$ | （て）$\downarrow$－ | （z）$\varsigma-z$ | （乙）$\varepsilon-1$ | （乙）$\varepsilon-1$ | （I）$\varepsilon-I$ | （ع）$t-I$ | 1 | I | （ع）$\downarrow-\boldsymbol{Z}$ | （SI） $8 \mathrm{I}-8$ | II |
| （I）$て$＇I | ${ }^{\prime} \mathrm{S}-2$ | （ $t$ ）$s-\tau$ | （ع）$t-1$ | （z） $\mathcal{E}-\mathrm{I}$ | （z）$て$＇I | （z）$\varepsilon-1$ | （ع）$t-I$ | （I）て＇I | （8）$\downarrow \mathrm{I}-\mathcal{E}$ | （I） $\mathrm{Z}^{\text {＇I }}$ | （I） $\mathcal{E}-\mathrm{I}$ | （て）て＇I | 01 |
| （乙） $\mathcal{E}$＇乙 | ${ }^{\text {S }}$－9 | （ع）9－ح | （b）$s-\varepsilon$ | （t）$\varsigma-\varepsilon$ | （b）$\varsigma-\varepsilon$ | （9） $0 \mathrm{I}^{-\mathrm{S}}$ | （6） $11-L$ | （S） $6^{--}$ | （8）$\varepsilon I-9$ | （9） 0 Iーマ | （6）$て I-S$ | （t） $9-z$ | 6 |
| （t）$L-\tau$ | ＇S－乙 | （乙）$t-\tau$ | （乙）$\varepsilon-I$ | （乙）$\varepsilon-I$ | （z）$\varepsilon-1$ | （乙）$\varepsilon$＇$\tau$ | （て）$\downarrow$－ |  | （6I）Sて－II | （SI） 0 O－8 |  | （を）$\downarrow-I$ | 8 |
| （5）9－t | ＇S－I | （ع）$s-\tau$ | （£）$\nabla^{\prime} \varepsilon$ | （ع）$\varsigma-\varepsilon$ | （t） $9-\varepsilon$ | （ $\downarrow$ ）$L-\varepsilon$ | （0て）$て$（て－てI | （0乙）$て$（ | （8I）Sて－SI | （b） $9-\varepsilon$ | （EI） $\mathrm{LI}-$ ZI | （てI）9I－0I | $L$ |
| － |  | （b） $9-\varepsilon$ | （ع）$\varsigma-\mathcal{E}$ | （¢）$\varsigma-\varepsilon$ | （S）L－ヤ |  | （LI）Iて－SI | （6I）\＆て－ヤI | （ $\varepsilon$ ）$t-Z$ | （ع） $8-\mathcal{E}$ | I | （0I）てI－8 | 9 |
| － | （b）$\varsigma-\varepsilon$ | （S） $8^{-t}$ | （9）$L$－ | （¢） $9-\mathcal{E}$ | （ $\downarrow$ ）$s-\varepsilon$ | （¢）9－ヤ | （9）8－9 | （ $\dagger$ ）$L-\varepsilon$ | してーロI | I | （દて） 2 ¢－8I | （6） $\mathcal{E I}-L$ | 5 |
| II－ナ | （z）$\varepsilon$＇$\tau$ | （I） $1 \times 1$ | 1 | （ع）$b-1$ | （ع）$\downarrow$－ | （c）$s-z$ | （t） $9-\varepsilon$ | （t） $9-\varepsilon$ | （ع） $\mathcal{E}-\mathrm{I}$ | （E）$t-I$ | （91）6I－II | （I） $\mathrm{Z}^{\prime} \mathrm{I}$ | $t$ |
| （9） $6-5$ | （c）$L-\varepsilon$ | （1） $\mathcal{E}-1$ | I | （I）$て ' I$ | （\＆）$b-z$ | I | I | I | （I） $\mathcal{E}-\mathrm{I}$ | （I）て＇I | I | ＋（I） $\boldsymbol{v}^{-1}$ | $\varepsilon$ |
| （SI） 1 て－てI | （S） $8-2$ | （E） $\mathcal{E}-1$ | I | I | （I）$て$＇I | （c） $\mathrm{S}-1$ | （c）$L-\mathcal{E}$ | （\＆）$v-\tau$ | I | （I）${ }^{\prime}$＇I | （8） $0 \mathrm{I}-\mathrm{L}$ | ＊$L$－ | $\tau$ |
| （I） 2 ＇I | （乙）$\varepsilon-1$ | （S）$L \rightarrow \downarrow$ | （8） 6 － 5 | （8） $01-5$ | （8） 0 I－ヤ | （9） $8-5$ | （b） $9-\varepsilon$ | （ع）$\downarrow-\sim$ | （z） $\mathcal{E}-1$ | （とて） $0 \varepsilon-9 \mathrm{I}$ | （L） $8-5$ | 1 | I |
|  | I | I | 1 | I | I | 1 | I |  | － |  | I | I | 0 |
| X | IIIA | IIA | 11 | $\wedge$ | 11 | III | II | 1 | L | W | d | $\stackrel{\supset}{\text { реว }}$ | on elos |
| stuvusas reutuopqy xeroul x |  |  |  |  |  |  |  |  |  |  |  |  |  |




Fig. 2. Fourth-instar larva of Anopheles (Cellia) ainshamsi. A, Head, dorsal (left) and ventral (right) aspects of left side. B, Thorax and abdominal segments I-VI, dorsal (left) and ventral (right) aspects of left side. C, Abdominal segments VII-X, left side. A, antenna; C, cranium; P, prothorax; M, mesothorax; MANP, median accessory tergal plate; S, spiracular lobe; T, metathorax; TP, tergal plate; I-VIII, X $=$ abdominal segments IVIII and $X ; 1-15=$ setal numbers for specified areas, e.g., seta $5-C$. Scales in mm .
branches; 11-T minute, sometimes absent; 12-T usually triple, sometimes double, rarely single. Abdomen: Integument hyaline, smooth; tergal plates on segments I-VII, roughly triangular, small, width 0.1 or less diameter of segments, median accessory tergal plates on segments II-VI. Seta 1-I weakly developed, with normal branches, usually triple, occasionally double, rarely with 4 branches, 1-II-VII palmate, with relatively few leaflets, leaflets darkly pigmented distally, generally lanceolate but some with weakly developed shoulders and short filaments; 3-I-III, V, VI fairly long, single (3-V occasionally double), 3-IV usually triple (2-4), 3-VII frequently double or triple but most often single; 9-I,II inserted more or less mesal to seta 7; 2-II,III,VII branched, 2-IV-VI single (2-VI rarely double); 6-IV-VI well developed, with long branches arising well beyond the base of central rachis. Pecten plate moderately tanned, darker on anterior margin, with 1217 (15) spines, spines with denticles principally on dorsal margins, one to few minute spicules on ventral margins, longer spines usually at each end with few interspersed among shorter spines. Seta $1-S$ large but with only $4-6(5)$ branches; 2-S usually with 4 or 5 branches ( $2-7$ ). Saddle moderately tanned, short, dorsal length $0.24-0.30$ mm (mean 0.27 mm ), lateral margins irregular in outline. Seta 1-X inserted on margin of saddle, long, about twice length of saddle, single or double, more often single; 2X with 12-21(15) branches, most branches on dorsal side of rachis, relatively straight, with fine tapering tips; 3-X with relatively few (5-9, mode 6) mostly long, thick, slightly curved, apically hooked branches; 4-X (ventral brush) with 9 offset pairs of setae. Anal papillae very small, short, ba-cillus-shaped, length about 0.09 mm .

DNA sequence.-Specimens available for this study included the type series that comprises material collected in 1983 (see below) and a series of pinned adults taken from a laboratory colony maintained at Harvard University in 1982. Ten of these
specimens, including two paratypes, were used for DNA extraction. Unfortunately, PCR amplification of the extracted DNA was unsuccessful using primers for the nuclear ITS2 region and the mitochondrial COI gene. Specimens preserved explicitly for DNA studies are needed for the molecular characterization of An. ainshamsi.

Etymology.-The species is named in recognition of Ain Shams University, Cairo, Egypt, and its support of mosquito biological research and vector control.

Systematics.-Anopheles ainshamsi, originally identified as An. stephensi, was first discovered near Râs Ghârib on the Gulf of Suez coast in 1966 (Gad 1967). Larvae and a few adults reared from larvae were sent to Dr Peter Mattingly in London, who noted "marked differences" between these specimens and specimens of An. stephensi in the BMNH (Gad and Kamel 1967). Despite the apparent differences, the Egyptian specimens were added to the museum's collection (now the NHM collection) of An. stephensi. The specimens include seven larvae mounted on a single microscope slide, four pinned females, and a pinned male with dissected genitalia on a microscope slide. The microscope slides are labeled "An. (Cellia) stephensi/EGYPT/Shokeir/ near Ras Ghareb/xi:1966/A.M. Gad/From brackish swamp/near seashore, Red/Sea coast", and the pinned adults each bear a label inscribed with "EGYPT/Shokeir/near Ras Ghareb/xi:1966/Brackish swamp/near seashore". Thus, it seems that Mattingly did not seriously question Gad's identification of the species despite the differences he observed in the adult and larval stages. After quoting the differences noted by Mattingly, Gad and Kamel (1967) suggested that the "marked differences in the Egyptian material might indicate that the mosquito has existed for a long time in the area."

Gad (1967) may have used Mattingly and Knight's (1956) keys to the mosquitoes of Arabia (the Arabian Peninsula south of the Sinai, Jordan and Iraq, and adjacent islands)
to identify the Anopheles mosquito from the Gulf of Suez coast. Adult females and larvae of An. ainshamsi both key to An. stephensi in these keys. Although the two species are similar, they are easily distinguished by the characters listed in Table 3. Some of these characters are illustrated and used to distinguish adult females of the two species in Glick's (1992) pictorial key to the anopheline mosquitoes of southwestern Asia and Egypt.

Females of An. ainshamsi lead to An. dancalicus in the pictorial key to the anopheline mosquitoes of Ethiopia constructed by Verrone (1962a), and are also identified as this species in the computer key to the Anopheles of the Afrotropical Region developed by Hervy et al. (1998). Females key to couplets that distinguish An. salbaii and An. dancalicus, and are identified as An. salbaii, in the keys to the Afrotropical anophelines by Gillies and de Meillon (1968) and Gillies and Coetzee (1987). It should be borne in mind, however, that $A n$. hervyi is not included in the last two keys because it was unknown when these keys were developed, and An. salbaii is not included in Verrone's key because it is not known to occur in Ethiopia. Hervy et al. (1998) included An. hervyi in their computer key even though it was not formally described until the following year.

Larvae of An. ainshamsi key to An. dancalicus in the Verrone's (1962b) pictorial key to the anopheline larvae of Ethiopia. They also key to this species in the keys of Gillies and de Meillon (1968) and Gillies and Coetzee (1987), but they are not identifiable as either An. dancalicus or An. salbaii in the computer key of Hervy et al. (1998). As in the case of Verrone's key for adult females, his key for larvae does not include An. salbaii. Unfortunately, the immature stages of An. hervyi are unknown.

Pupae fail to be identified as either An. dancalicus or An. salbaii in the keys of Gillies and de Meillon (1968) and Gillies and

Coetzee (1987), which are the only available keys for the identification of this life stage of Afrotropical Anopheles. Identification terminates at couplet 25 because seta $1-\mathrm{V}, \mathrm{VI}$ is long in An. ainshamsi and must be short to key to An. dancalicus and An. salbaii.

Anopheles ainshamsi obviously belongs to the Neocellia Series, a group of species that breed in open temporary pools of water and are characterized by the presence of broad scutal scales and the absence of upper proepisternal setae in the adults. Larvae have one long mesopleural and two long metapleural setae branched. The series, as currently defined, includes 16 species divided between three species groups and 14 species, including An. dancalicus, An. hervyi, An. salbaii and An. stephensi, that are unassigned to species groups (Harbach 2004). As An. ainshamsi does not exhibit features that diagnose any of the currently recognized species groups, and shares salient anatomical features with four unassigned species, it must be regarded as another unassigned species of the Neocellia Series. Based on overall morphological similarity, as indicated in Table 3 and reflected in the use of the identification keys mentioned above, An. ainshamsi appears to be more closely related to An. dancalicus than to the other three species. Areas of the scutum and wings of the adults bear the same ornamentation and markings, pupal setae $1-\mathrm{V}-$ VII and $7-$ VI,VII are similarly developed, and the leaflets of larval setae 1-IV-VII are lanceolate (usually) or have weakly developed shoulders and a short filament.

Bionomics.-Larvae of An. ainshamsi occur in shallow clear saltwater pools, usually shaded by halophilic shrubs, Avicennia marina (Forsk.) (Avicenniacae: Verbenaceae), and various grasses. The water sometimes contains dense mats of grass and filamentous green algae. Larvae are also abundant in depressions and drilled holes without vegetation. Larvae of Ochlerotatus




| Stage | Anatomeal Peature | An. ainshamsi | An. stephensi | An. dancalicus | An. salbaii | An. herryi |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| Pupate | Seta 9-IV | More or less peglike with rather blunt apex | Peglike with blunt apex | More or less spinelike with acute apex | Peglike with blunt apex | ? |
|  | Seta 9-V-VII | Long, slender. simple | Long, relatively stout, simple | Long, relatively slender, usually bi- or trifurcate | Long, stout, usually simple | ? |
|  | Seta 1-V-VII | Single, longer than tergum | Usually single, longer than tergum | Branched, not longer than tergum | Usually single and longer than tergum | $?$ |
|  | Seta 7-VI.VII | Long, about length of tergum, usually single | Short, less than half length of tergum, usually branched | Long, about length of tergum, usually single | Short. less than half length of tergum, single or branched | ? |
| Larvae | Seta 2-C | Aciculate or frayed | Often aciculate or frayed | Simple or aciculate | Simple | $?$ |
|  | Seta 3-C | Aciculate or frayed | Simple | Simple | Simple | ? |
|  | Seta 1-X | Single or double | Single | Single | Single | ? |
|  | Setae I-IV-VII | Branches lanceolate or with weak shoulders and short filament | Branches with distinct shoulders and filament | Branches lanceolate or with weak shoulders and short filament | Branches with distinct shoulders and filament | ? |



Fig. 3. Typical breeding site of Anopheles (Cellia) ainshamsi near Râs Shukeir on the Gulf of Suez coast.
detritus (Haliday) also occur in these habitats. The type series consists of larvae and specimens reared from larvae and pupae collected from salt encrusted pools and drilled holes ranging from $0.2-2.0 \mathrm{~m}$ in diameter. A typical saline pool inhabited by larvae is shown in Fig. 3.

The pH and salinity of breeding sites were recorded using methods recommended by the World Health Organization (WHO 1975). The pH of pools near Râs Ghârib ranged from 6.9-7.3; those that were not encrusted with salt contained 33-37.5 g $\mathrm{Cl}^{-}$/ liter whereas those encrusted with salt contained $54-72 \mathrm{~g} \mathrm{Cl}^{-} /$liter. Pools at the site north of Râs Shukeir were more saline and more acidic: $72-77.5 \mathrm{~g} \mathrm{Cl}^{-/}$liter, pH 5.8-6.3. However, An. ainshamsi develops well in water of various salinities. Gad et al. (1987) reared them successfully both in
seawater ( $21-22 \mathrm{~g} \mathrm{Cl}^{-} /$liter) and water with only $9 \mathrm{~g} \mathrm{Cl}^{-/}$liter.

Anopheles ainshamsi occurs in areas that are uninhabited (Riberio et al. 1985, Kenawy 1988), but females will attack humans who visit their realm (Gad and Salit 1972). Lizards, deer, and passerine birds have been seen near breeding places (Gad and Kemal 1967, Ribeiro et al. 1985), and camel skeletons were observed in the salt springs area, but whether females feed on these or other animals is unknown. As noted previously, An. ainshamsi is an autogenous mosquito capable of developing and laying eggs in the absence of hosts (Ribeiro et al. 1985).

Distribution.-Anopheles ainshamsi is known only from coastal areas of Râs Shukeir District, El-Bahr El-Ahmar (Red Sea) Governorate, Egypt. Larvae have been collected from sites not far from the Gulf of Suez coast. One site, where this species was originally discovered and identified as $A n$. stephensi by Gad (1967), is located near Râs Ghârib. Another site is located about 19 km north of Râs Shukeir town, and the specimens that comprise the type series include fourth-instar larvae and adults reared from larvae and pupae collected about 5 km west of Râs Shukeir.

Type series.-Two hundred and eighteen specimens ( 2 adults used for DNA extraction; failed PCR): 32 ㅇ, 33 đ, 4 oे genitalia, $40 \mathrm{Le}, 65 \mathrm{Pe}, 44 \mathrm{~L}$. Holotype, $\frac{q}{}$ (EG146-34), with LePe on microscope slide, EGYPT: El-Bahr El-Ahmar (Red Sea) Governorate, 5 km W of Râs Shukeir, mixed small ground pools and holes, some drying up and encrusted with salt, 30 April 1983 (Harrison, Gad, Gamal). Paratypes (same locality and collectors as holotype), 25 \& LePe (EG146-2, -4, -5, -6, -9, -11, $-12,-18,-23,-25$ through -40 ; EG146-23 used for DNA extraction, failed PCR); 14 ठ LePe (EG146-1, $-3,-7,-8,-10,-13$ through -17, -19 through -22, -24; EG146-$1,-3,-7,-8$ with dissected genitalia on microscope slides; EG146-14 used for DNA extraction, failed PCR); 7 ¢ Pe (EG146-100
through -103, -106, -109, -121); 18 ơ Pe (EG146-104, -105, -107, -108, - 111 through -120, -122, -123, -124); 44 L (EG146). The holotype (EG146-34) and the following paratypes are deposited in the NMNH: EG146-1, $-2,-3,-6$ to $-9,-11$ to $-16,-18$ to $-22,-24$ to -40 , and 40 L . The remaining paratypes are deposited in Ain Shams University (EG146-4, -17, -103, -104, and 2L) and the NHM (EG146-5. $-10,-14,-23,-109,-122$, and 2L).

## Acknowledgments

We are grateful to Ali Hafez and Saad El Masry for assistance in the field in Egypt; to Taina Litwak (formerly of WRBU) for preparing pencil drawings of the larva and pupa; to Theresa Howard (NHM) for modifying and inking the pencil drawings and illustrating of male genitalia; to Joanna Elphic (formerly of NHM) for recording larval and pupal chaetotaxy; and to Yvonne Linton and Fredy Ruiz (NHM) for conducting DNA extractions and PCR in an attempt to obtain DNA sequences from available specimens.

## Literature Cited

Brunhes, J., G. le Goff, and B. Geoffroy. 1999. Afrotropical anophelines mosquitoes. III. Description of three new species: Anopheles carnevalei sp. nov., An. hervyi sp. nov., and An. dualaensis sp. nov., and resurrection of An. rageaui Mattingly and Adam. Journal of the American Mosquito Control Association 15: 552-558.
El-Said, S. and M. Kenawy. 1983. Geographical distribution of mosquitoes in Egypt. Journal of the Egyptian Public Health Association 58: 46-76.
Gad, A. M. 1967. Anopheles stephensi Liston in Egypt, UAR. Mosquito News 27: 171-174.
Gad, A. M. and O. M. Kamel. 1967. Further notes on Anopheles stephensi in Egypt, U.A.R. Journal of the Egyptian Public Health Association 42: 249252.

Gad, A. M. and A. Salit. 1972. The mosquitoes of the Red Sea area, Egypt. Journal of Medical Entomology 9: 581-582.
Gad, A. M., S. El Said, A. N. Hassan, and A. Shoukry. 1987. The distribution and ecology of the mosquitoes in the Red Sea Governorate, Egypt. Journal of the Egyptian Society of Parasitology 17: 207-221.
Gad. A. M.. W. A. Maier, and G. Piekarski. 1979a.

Pathology of Anopheles stephensi after infection with Plasmodium berghei berghei. I. Mortality rate. Zeitschrift fü Parasitenkunde 50: 249-261.

1979b. Pathology of Anopheles stephensi after infection with Plasmodium berghei berghei. II. Changes in amino acid contents. Zeitschrift für Parasitenkunde 60: 263-275.
Gillies, M. T. and M. Coetzee. 1987. A supplement to the Anophelinae of Africa south of the Sahara (Afrotropical Region). Publications of the South African Institute for Medical Research 55: 1143.

Gillies, M. T. and B. de Meillon. 1968. The Anophelinae of Africa south of the Sahara (Ethiopian zoogeographical region). 2nd ed.. Publications of the South African Institute for Medical Research 54: 1-343.
Glick, J. I. 1992. Illustrated key to the female Anopheles of southwestern Asia and Egypt (Diptera: Culicidae). Mosquito Systematics 24: 125-153.
Harbach, R. E. 2004. The classification of genus Anopheles (Diptera: Culicidae): a working hypothesis of phylogenetic relationships. Bulletin of Entomological Research 94: 537-553.
Harbach, R. E. and K. L. Knight. 1980. Taxonomists' Glossary of Mosquito Anatomy. Plexus Publishing, Inc., Marlton, New Jersey. xi +415 pp.
——. 1982 [1981]. Corrections and additions to Taxonomists' Glossary of Mosquito Anatomy: Mosquito Systematics 13: 201-217.
Hervy, J.-P., G. le Goff, B. Geoffroy, L. Hervé, J.-P. Manga, L. Manga, and J. Brunhes. 1998. Les anophèles de la région afro-tropicale. CD-ROM. ORSTOM, Montpellier, France.
Kenawy, M. A. 1988. Anopheline mosquitoes (Diptera: Culicidae) as malaria carriers in A.R. Egypt "history and present status". The Journal of the Egyptian Public Health Association 63: 67-85.

- 1990. Fauna of anopheline mosquitoes (Diptera: Culicidae) in A.R. Egypt "historical background and present situation". The Journal of the Egyptian Public Health Association 65: 263281.

Mattingly, P. F. and K. L. Knight. 1956. The mosquitoes of Arabia I. Bulletin of the British Museum (Natural History) Entomology 4: 91-141.
Ribeiro, J. M. C., P. A. Rossignol, and A. Spielman. 1985. Salivary gland apyrase determines probing time in anopheline mosquitoes. Journal of Insect Physiology 31: 689-692.
Verrone, G. A. 1962a. Outline for the determination of malarial mosquitoes in Ethiopia. Part I-adult female anophelines. Mosquito News 22: 37-49.
. 1962b. Outline for the determination of malaria mosquitoes in Ethiopia. Part II- anopheline larvae. Mosquito News 22: 394-401.
Ward, R. A. 1992. Third supplement to "A Catalog of
the Mosquitoes of the World" (Diptera: Culicidae). Mosquito Systematics 24: 177-230.
WHO. 1975. Manual on Practical Entomology in Malaria. Part II. Geneva, WHO offset publication, no. 13. 191 pp .

Wilkerson, R. C. and E. L. Peyton. 1990. Standardized nomenclature for the costal wing spots of the genus Anopheles and other spotted-wing mosquitoes (Diptera: Culicidae). Journal of Medical Entomology 27: 207-224.

